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ECOLOGY

Avoiding (Re)extinction

Alternative methods of identification should be used to avoid collection of voucher specimens of threatened or rediscovered species.

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Field biologists have traditionally collected voucher specimens to confirm a species' existence. This practice continues to this day but can magnify the extinction risk for small and often isolated populations. The availability of adequate alternative methods of documentation, including high-resolution photography, audio recording, and nonlethal sampling, provide an opportunity to revisit and reconsider field collection practices and policies.

Cases such as the extinction of the great auk remind us what is at stake in taking animals from small and declining populations. The last wild great auk (*Pinguinus impennis*) was sighted in 1844 on Eldey Island, Iceland. Centuries of exploitation for food and feathers, and, to some degree, a changing climate, had stressed the species, but overzealous museum collectors also played a role in its extinction (*1*). As the bird's numbers dwindled in the 19th century, ornithologists and curators increasingly prized great auk skins and eggs, with museums and universities sending out collection parties to procure specimens. On Eldey, fishermen killed the final breeding pair of the flightless birds and sold them to a local chemist, who stuffed the specimens and preserved them in spirits. Their internal organs now reside at the Zoological Museum in Copenhagen (*2*).

The great auk's disappearance predates the rise of a robust societal ethic of conservation and the emergence of a scientific concern for global biodiversity decline in the late 20th century. Yet, there is still a strong and widespread impulse to procure specimens of rare or rediscovered species for scientific purposes.

In their global review of species reappearances, Scheffers *et al*. (*3*) document at least 351 species that have been rediscovered since 1889, mostly in the tropics. In recent years, scientific and media attention has been drawn to the rediscovery of amphibian species thought to be extinct, including 11 species in Costa Rica alone (see the figure). Many amphibian rediscoveries have been documented by collecting specimens upon first encounter, a practice one of us has carried out in the past [R.P. with *Craugastor ranoides*, (*4*)]. Such rediscovered species typically exist in small populations with small range sizes and are therefore highly vulnerable. The desire to collect voucher specimens to verify the reappearance of species presumed extinct can be heightened by the recognition of the organism's rarity, as in the case of private individuals seeking to own and display rare animal specimens for their perceived scarcity and thus value. Rediscoveries can also be accidental, as many missing species are hard to identify in the field and collected specimens may turn out to be from very small populations, with the risk of collection only realized well after the fact (*5*).

PHOTO CREDIT: JUAN G. ABARCA ALVARADO

Species loss and rediscovery in Costa Rica. The fungal pathogen *Batrachochytrium dendrobatidis* (Bd) has been linked to the decline and extinction of amphibians worldwide (*12*). For example, amphibian populations in Costa Rica experienced substantial declines, with 20 of the 199 species feared extinct, after Bd moved through the country from the mid-1980s to the early 1990s (*13*, *14*). However, 11 of the 23 species have been rediscovered (*4*). Holdridge's toad (*Incilus holdridgei*) (see photo), a species endemic to a single volcano, vanished during the declines and was declared extinct by the International Union for Conservation of Nature and Natural Resources in 2007 but was rediscovered in 2008. Today, relict populations persist in areas where Bd once contributed to their demise.

Many taxa are difficult to identify from morphology alone. The collection of voucher specimens by field biologists is therefore increasingly augmented by other kinds of samples. Cultural traditions within a research community can, however, reinforce the collection of voucher specimens even where it is not necessary by insisting that a preserved specimen in a natural history collection is the gold standard—or only standard—for publishing a species description or documenting a species' presence. Collecting specimens is no longer required to describe a species or to document its rediscovery.

The concern about overcollection goes well beyond the case of rediscovered spe-

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cies. It also applies to the more common scenario of documenting newly discovered species, which (like most rediscovered species) often exist in small, isolated populations and therefore suffer from the same problems if voucher specimens are collected from the field. Field collection of individuals from small and declining populations vulnerable to extinction is also a common practice. Collection both by professional and amateur scientists has been linked to the decline or loss of a range of animal species, including Mexico's elf owl (*Micrathene whitneyi soccorroensis*) (*6*). Plants have also been affected by scientific overcollection; Norton *et al*. (*7*) cite the case of the scientific collection–driven decline and extinction of uncommon plant taxa in New Zealand over the past two centuries.

Perhaps the most powerful alternative method to collection is a series of good photographs, which can even be used to describe a species, complemented by other lines of evidence, such as molecular data and a description of a species' mating call for birds, amphibians, or insects. Advances in handheld technology have made it much easier and cheaper to identify species; most smartphones have a camera and a voice recorder sufficient to gather high-resolution images as well as an organism's call. Such nonlethal techniques were used successfully for the identification of the bird *Bugun liocichla*, a species that was newly discovered in India in 2006 (*8*). The bird's discoverer deliberately chose not to collect a voucher specimen for fear of imperiling the population; instead, a combination of photos, audio recordings, and feathers were used to distinguish the species.

In the case of rediscovered species, many were already well described, and a good-quality image should suffice. For rediscovered, rare, and newly discovered species, molecular techniques (such as skin swabbing for DNA) are an increasingly effective way to sample a specimen to confirm an identity with no or minimal harm to the organism (*9*, *10*). For this system to work, the DNA of relict populations and newly discovered species must be sequenced and the data made publicly available. This would, for example, make future population rediscoveries easier to document.

The multivariate description of a species that results from combining high-resolution photographs, sonograms (as appropriate), molecular samples, and other characteristics that do not require taking a specimen from the wild can be just as accurate as the collection of a voucher specimen without increasing the extinction risk. Clearly there remains a long-running debate over the appropriate standards for scientific description absent a voucher specimen (*11*). The benefits and costs of verification-driven specimen collection, however, should be more openly and systematically addressed by scientific societies, volunteer naturalist groups, and museums. Sharing of specimen information, including obligations to store genetic information from voucher specimens in widely accessible digital repositories, can also help to reduce the future need to collect animals from the wild.

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MATERIALS SCIENCE

Exploring the Interface of Graphene and Biology

To take advantage of the properties of graphene in biomedical applications, well-defined materials need to be matched with intended applications.

Kostas Kostarelos[1,3] and Kostya S. Novoselov[2,3]

Graphene is highly conductive, flexible, and has controllable permittivity and hydrophilicity, among its other distinctive properties (*1*, *2*). These properties could enable the development of multifunctional biomedical devices (*3*). A key issue for such applications is the determination of the possible interactions with components of the biological milieu to reveal the opportunities offered and the limitations posed. As with any other nanomaterial, biological studies of graphene should be performed with very specific, well-designed, and well-characterized types of materials with defined exposure. We outline three layers of complexity that are interconnected and need to be considered carefully in the development of graphene for use in biomedical applications: material characteristics; interactions with biological components (tissues, cells, and proteins); and biological activity outcomes.

Graphene has now been developed in many different forms in terms of shapes, sizes, chemical modifications, and other characteristics that can produce dramatically different results when studied biologically. Methods for producing graphene include direct exfoliation in organic liquids (*4*, *5*), reduction of graphene oxide (GO) (*6*), and epitaxial growth by CVD (chemical vapor deposition) on copper (*7*) or epitaxial growth on silicon carbide (*8*). The three aspects of this layer of structural complexity—the thickness, the lateral extent, and the surface functionalization of graphene—are illustrated in panel A of the figure and show how the materials produced by different methods fall in very different parts of this parameter space. These different physical and chemical characteristics dictate the suitability of a material for specific biomedical applications.

These wide discrepancies between the available graphene types will crucially determine the second layer of complexity, that of interactions of graphene with living cells and their compartments. In panel B

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